

## EFFECT OF CROP COVER AND STAGE OF CROP GROWTH ON SOIL L-ASPARAGINASE, ALKALINE AND ACID PHOSPHATASE IN VERTISOL

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### ABSTRACT

*To emphasize the importance vegetation type and the age of vegetation a pot culture experiment was designed and conducted on Vertisol, in the greenhouse of Department of Soil Science and Agricultural Chemistry, Rajendranagar, Hyderabad, Telangana state, India. To study the influence of crop cover and stage of crop growth on soil enzyme L-asparaginase activity in soil. The experiment was undertaken with three cereals – rice, sorghum and maize, two oil seeds – groundnut and sesame, two pulses – green gram and black gram, two vegetables – bhendi and brinjal. The experiment was conducted using crops as treatments in Completely Randomized Block design with three replications along with the uncropped control. The results obtained with regard to the effect of these crops soil L-asparaginase activity showed that there was an increase in enzyme activity with age of the crop and it varied with plant species grown. The enzyme activity increased from 0 days and reached pick level at 60 days for l-asparaginase and 45 days for acid and alkaline phosphatases. The increase in L-asparaginase activity (expressed as  $\mu\text{g}$  of  $\text{NH}_4^+$  released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ ) ranged from 1.90 to 7.11 in groundnut (*Archishypogaea*), from 1.88 to 4.54 in black gram (*Vignamungo*), from 1.88 to 4.38 in - green gram (*Vignarabiata*), from 1.88 to 4.25 in sesame (*Sesamumindicum*), from 1.86 to 4.16 in rice (*Oryza sativa*), 1.86 to 3.96 in maize (*Zea mays*), from 1.86 to 3.82 in sorghum (*Sorghum vulgare*), from 1.85 to 3.12 in brinjal (*Solanummelongena*) and from 1.85 to 3.05 in bhendi (*Abelmoschusesculentus*). The activity of L-asparaginase, acid and alkaline phosphatase under different crop coverages followed the order groundnut >blackgram>greengram> sesame > rice > maize > sorghum >brinjal>bhendi. The presence and type of plants grown on a soil have shown a marked influence on enzyme activities, more over the levels of enzymes activity declined to nearly the original levels at harvest.*

**KEYWORDS:** L- Asparaginase, Cropcover, Acid and Alkaline Phosphatase

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### INTRODUCTION

The enzyme L-asparaginase (L-asparagine amidohydrolase EC 3.5.1.1) plays an important role in N mineralization of soils. The chemical nature of N in soils is such that a large proportion (15 -25%) of the total soil N is often released as  $\text{NH}_4^+$  by acid hydrolysis (6N of HCL). (Sowden, 1958) suggested that a portion of the released  $\text{NH}_4^+$  comes from the hydrolysis of amide (asparagine and glutamine) residues in soil organic matter. Bremner (1955) reported that hydrolysis of humic preparation released 7.3 to 12.6% of total nitrogen in the form of amide nitrogen. Sowden (1958) also reported that a percentage of the  $\text{NH}_4^+$  released during acid hydrolysis was equal to or nearly equal to the sum of nitrogen released from asparatic acid – N plus glutamic acid – N derived from of

asparagines and glutamine. Studies to understand the role of L-asparaginase, in soil N cycling and the factors (i.e., soil properties, and crop cover and stage of crop, nutrient management practices) that affect the activity of this enzyme will aid in fertility, productivity and sustainability of soils.

The presence of crop cover and the type of plant grown on the soil will have marked effect on the enzyme activities. Their effect could be either direct through endoenzymes contained in the plant residues, the extracellular enzymes secreted by living roots may also make significant contribution to their activity. The type of crops grown and the stage of Crops contribute to the enzyme pool in the soil either through enzyme exudation or indirectly through exudation of carbon substrates which promote microbial growth (Dick, 1994). This is because cropping systems that have higher carbon input or that conserve carbon inputs promotes enzyme activity. The higher organic matter input resulted in greater soil aggregation.

It is well known that soil quality and fertility is related to the biological activity of a soil, a key factor in organic systems (Stockdale & Watson 2009). Soil enzyme production as a result of microbial metabolism is a sensitive indicator of soil microbial activity, reflecting the release of nutrients for plant and microorganism growth. In addition, enzyme activity responds to agricultural practices involving, for example, fertilizers, organic amendments, tillage, cover crops and pesticides (Gianfreda & Bollag 1996). Some authors have indicated that the use of cover crops and tillage can alter enzymatic activity (Mikanova et al. 2006; Emmerling 2007). In fact, the amount of research available on the influence of vegetation species and the age of vegetation on soil enzyme is sketchy, not much information is available on L-asparaginase, therefore the present study was undertaken. These enzymes were selected as they plays a major role in N transformation and involved in mineralization of organically bound phosphorous to inorganic phosphorus compounds in soil and are the most important in plant nutrition.

## MATERIALS AND METHODS

A pot culture experiment was conducted on Vertisol, in the green house of the Department of Soil science and Agricultural chemistry, College of Agriculture, Rajendranagar, Hyderabad during the year 2014. The experiment was undertaken with three cereals –rice (*Oryza sativa*) sorghum(*Sorghum vulgare*) and maize (*Zea mays*), two oil seeds – groundnut (*Archishypogaea*) and sesame (*Sesamum indicum*), two vegetables – bhendi (*Abelmoschus esculentus*) and brinjal (*Solanum melongena*) and two pulses - green gram (*Vignarabiata*) and black gram (*Vignamungo*). The experiment was conducted with crops as treatments in Completely Randomized Block design with three replications along with uncropped control. The crops received recommended agricultural practices during crop growth. The initial soil sample was taken on the date of sowing and subsequence samples were collected at fifteen days interval till harvest. The effect of crop cover and stage of crop growth on L-asparaginase the activity of asparaginase was estimated by steam distillation method by estimating  $\mu\text{g}$  of  $\text{NH}_4^+$  released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$  by (Frankenberger and Tabatabai, 1991a) acid phosphatase (Tabatabai and Bremner, 1969) and alkaline phosphatase (Eivazi and Tabatabai, 1977) activity were studied by estimating  $\mu\text{g}$  of 4-nitrophenol released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ .

## RESULTS AND DISCUSSIONS

The results obtained with regard to the effect of crop cover and stage of crop growth on L-asparaginase activity for soil are presented in table 1. L-asparaginase activity (expressed as  $\mu\text{g}$  of  $\text{NH}_4^+$  released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ ) in soils collected under different crops varied with the crops grown The enzyme activity was consistently higher in soils covered with

groundnut, blackgram, greengram, sesame, rice, maize, sorghum, brinjal and bhendi. The activity of L-asparaginase increased with the age of crop and increase recorded at each stage of crop growth was significantly higher over previous crop stage upto 60 days after sowing and then steadily decreased till harvest. The increase in enzyme activity varied in groundnut from 1.90 to 7.11, black gram from 1.88 to 4.54, green gram from 1.88 to 4.38, sesame from 1.88 to 4.25, rice from 1.86 to 4.16, maize 1.86 to 3.96, sorghum from 1.86 to 3.82, brinjal from 1.85 to 3.12 and bhendi from 1.85 to 3.05  $\mu\text{g}$  of  $\text{NH}_4^+$  released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ . The activity of L-asparaginase in soil under different crop covers followed the order groundnut > blackgram > greengram > sesame > rice > maize > sorghum > brinjal > bhendi.

The acid phosphatase activity (expressed as  $\mu\text{g}$  of 4- nitrophenol released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ ) value for groundnut ranged from 15.26 to 54.89, black gram from 15.23 to 50.34, green gram from 15.23 to 49.71, sesame from 15.23 to 48.77, rice from 15.23 to 50.95, maize 15.22 to 47.92, sorghum from 15.22 to 46.98, brinjal from 15.21 to 40.51 and bhendi from 15.21 to 40.23 ( figure 1) enzyme activity in the soil under different crop coverages followed the same order as that for L-asparaginase. For the plant species grown, the soil acid phosphatases activity increased with the age of the crop upto 45 days except for rice the increased was observed upto 60 days. For the growth period from 0 to 105 DAS, the acid phosphatase activity was found to be significantly higher than their corresponding controls for all the crop plants.

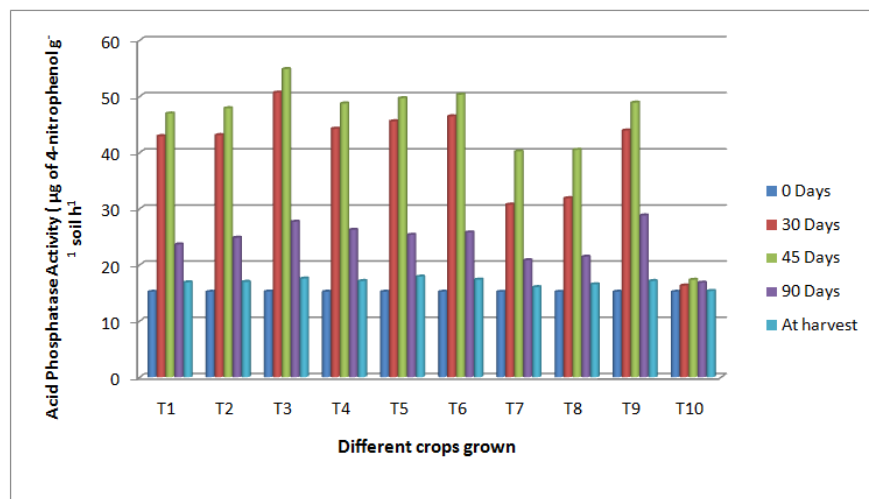
A close persual of the data indicated that the value of alkaline phosphatase (expressed as  $\mu\text{g}$  of 4- nitrophenol released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$ ) in uncropped control ranged from 33.27 to 39.95 in soil. The activity increased sharply upto 60 days after transplanting in rice and upto 45 days in other crops after sowing and there after the activity gradually decreased gradually to nearly 0 day level for all the cropped pots. The highest alkaline phosphatase activity was found under groundnut cover which ranged from 33.39 to 65.55 followed by black gram that ranged from 33.38 to 62.18, greengram from 33.37 to 61.37, sesame 33.29 to 60.70, rice from 33.37 to 61.75, maize 33.30 to 59.77, sorghum from 33.30 to 59.50, brinjal from 33.31 to 55.25 and bhendi from 33.32 to 56.69. These results are in conformity with the trends reported in literature by various workers (Vandana *et al.*, 2012; Hamido *et al.*, 2009 and Ting *et al.*, 2015)

**Table 1: Effect of Crop Cover on Soil L-Asparaginase Activity ( $\mu\text{g}$  of  $\text{NH}_4^+$  - N Released  $\text{G}^{-1}$  Soil  $\text{H}^{-2}$ )**

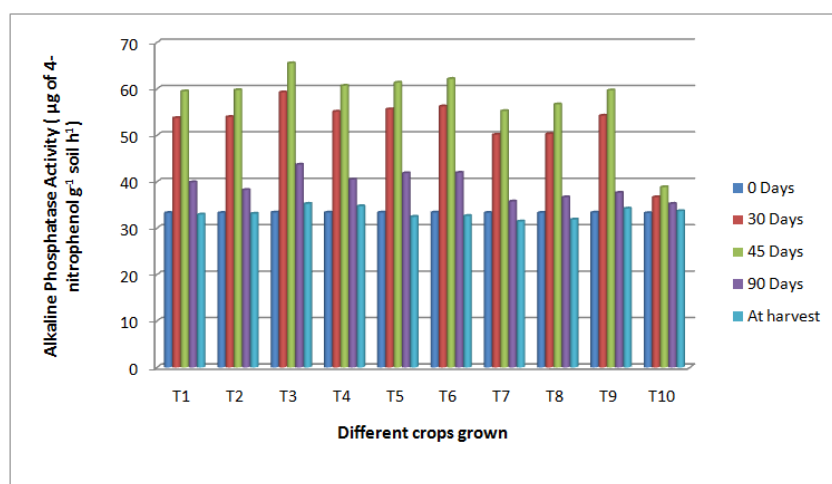
Treatments	Days After Sowing								
	0	15	30	45	60	75	90	105	Mean
Sorghum	1.86	2.33	2.66	2.96	3.82	3.15	2.36	1.95	2.64
Maize	1.86	2.36	2.74	3.08	3.96	3.21	2.44	2.04	2.71
Groundnut	1.90	3.10	4.24	5.93	7.11	5.61	3.48	2.13	4.19
Sesame	1.88	2.63	2.93	3.56	4.25	3.46	2.56	2.04	2.92
Green gram	1.88	2.76	3.13	3.82	4.38	3.57	2.68	2.10	3.04
Black gram	1.88	2.83	3.28	3.94	4.54	3.69	2.79	2.12	3.13
Brinjal	1.85	2.14	2.41	2.75	3.05	2.44	2.18	1.85	2.33
Bhendi	1.85	2.21	2.48	2.87	3.12	2.52	2.25	1.94	2.41
Rice	1.86	2.44	3.05	3.26	4.16	3.34	2.85	2.24	2.82
Control	1.84	2.01	2.43	2.72	2.96	2.83	2.56	1.90	2.41
Mean	1.87	2.48	2.93	3.49	4.14	3.38	2.62	2.03	2.86

**Table 2**

Analysis of Variance	C.D At 5 %	Sem $\pm$
Plant cover	0.02822	0.0101
Days after sowing	0.02524	0.0090
Plant Cover x Days after Sowing	0.07982	0.0285



**Figure 1: Effect of Crop Cover on Soil Acid Phosphatase Activity ( $\mu\text{g}$  of 4-Nitrophenol Released  $\text{G}^{-1}$  Soil  $\text{H}^{-1}$ )**



**Figure 2: Effect of Crop Cover on Soil Alkaline Phosphatase Activity ( $\mu\text{g}$  of 4-Nitrophenol Released  $\text{G}^{-1}$  Soil  $\text{H}^{-1}$ )**

Soil enzymes including l-asparaginase and phosphatase which were studied are generally activated to various degrees by the vegetative sites than that of control, this is due to the cropped sites have higher organic matter input to soil, which inturn increases the carbon turnover and there by availability of nutrients. Hence enzyme activity depends on the addition of crop residue; to emphasize the importance of Nitrogen substrates by legumes they were also included in the study.

Vandana *et al* (2012) reported that the acid phosphatase activity ranged from 12.84 to 51.59  $\mu\text{g}$  *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{h}^{-1}$  in under cover crops and 12.34 to 19.32  $\mu\text{g}$  *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{h}^{-1}$  in control. The quality and quantity of litter play a major role on the growth of soil microbial community. Soil mineralization processes differ among the plant species. These differences could be due the combined effects of the quality of soil substrate, physico-chemical properties and inturn of microbial composition. Acid phosphatase activity was found by many workers to be positively correlated with organic C, soil moisture, total N, and C/N ratio, which inturn depend on microbial load of soil, indicating that microbial biomass increased with increasing soil organic C and total N. Therefore, high acid phosphatase activity may partly result from a high microbial biomass. As stated by Venterink(2011) high acid phosphomonoesterase activity in soil under legumes was linked to the phosphatase activity of roots rather than to soil microbial activity. According to

(Pantelista *et al* 2012) the ratio of soil phosphomonoesterase activity to microbial biomass increased significantly during flowering period of the plants, probably indicating over synthesis of this enzyme by microbes at this period this can be attributed both to roots and microbial phosphatase activity. Similar temporal pattern to that of phosphomonoesterase was also exhibited by glutaminase, during flowering the root nodules where intense N fixation occurs were well developed. Hence it is possible that fixed  $\text{NH}_4^+$  is converted into n-rich compounds including glutamine and glutamic acid and this may have stimulate glutaminase activity of soil once vegetable residues reach soil Hamido and Kpomblekou (2009)

In a study conducted by (Tang *et al* 2014) the alkaline phosphatase activity of different soils collected at different growth stages was in the range of 108.34–186.37 and 92.02–169.12  $\mu\text{g } p\text{-nitrophenol g}^{-1} \text{ soil h}^{-1}$ , respectively. The highest activities were detected at the booting stage. The different ranking of treatments in soil enzyme activities might be related to the kinds of winter cover crops straw type. This might have been because there was significant difference decomposable organic material in the winter cover crops straw-returned soil which favored soil enzyme activities by increasing the number of microbial load.

Balota *et al* (2011) reported that variation of soil enzyme activities are due to soil tillage system and different ground cover crops, these enzymes were sensitive to soil disturbance hence more enzyme activity was found in the grass interrows than the main crop (legumes) this is due to the production of more biomass, which indirectly plays a major role in microbial growth and thus enzyme production.

## CONCLUSIONS

In a study conducted by (Hamido and Kpomblekou 2009) plots preceded by Crimson clover showed higher activity than black oats and a mixed plot showed even more activity of L-asparaginase activity this is due to decomposition of crimson clover foliage and root biomass which enhance the activity it also showed that the enzyme activity was more in cover plot than control plots. In the present study activities of these enzymes aryl amidases, l-glutaminases l-asparaginase and ureases varied with cover crops and soil depth.

All the soil enzymes are significantly correlated with organic matter content of the soil. in the soil under cover crops higher enzyme activity was reported this is due to stimulation of microbial biomass by the addition of repeated incorporation of organic residues, enhanced humus content, accumulation/stabilization of organic matter, abundance of carbohydrates coupled with greater microbial activities

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